

(12)特許協力条約に基づいて公開された国際出願。

(19) 世界知的所有権機関
国際事務局



(43) 国際公開日
2004 年 10 月 28 日 (28.10.2004)

PCT

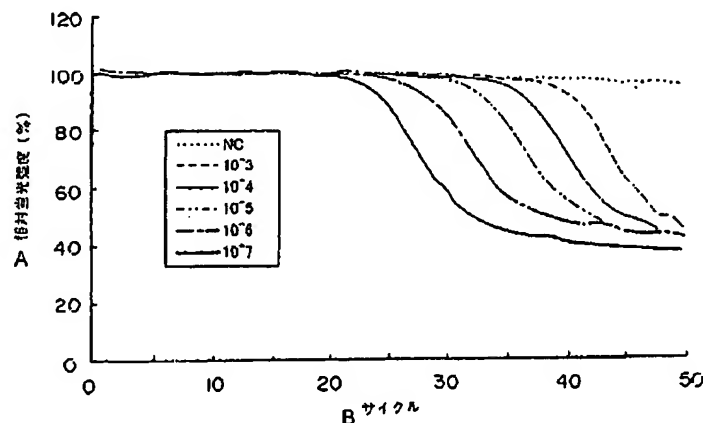
(10) 国際公開番号
WO 2004/092415 A1

- (51) 国際特許分類: C12Q 1/68, C12N 15/09 (72) 発明者; および
(21) 国際出願番号: PCT/JP2004/005496 (75) 発明者/出願人 (米国についてのみ): 平井 光春 (HIRAI, Mitsuharu) [JP/JP]; 〒6018045 京都府京都市南区東九条西明田町 5 7 番地 アークレイ株式会社内 Kyoto (JP).
(22) 国際出願日: 2004 年 4 月 16 日 (16.04.2004)
(25) 国際出願の言語: 日本語 (74) 代理人: 川口 嘉之, 外 (KAWAGUCHI, Yoshiyuki et al.); 〒1030004 東京都中央区東日本橋 3 丁目 4 番 10 号 アクロポリス 2 1 ビル 6 階 Tokyo (JP).
(26) 国際公開の言語: 日本語
(30) 優先権データ:
特願 2003-111173 2003 年 4 月 16 日 (16.04.2003) JP
特願 2003-114382 2003 年 4 月 18 日 (18.04.2003) JP
(71) 出願人 (米国を除く全ての指定国について): アークレイ株式会社 (ARKRAY, INC.) [JP/JP]; 〒6018045 京都府京都市南区東九条西明田町 5 7 番地 Kyoto (JP).
(81) 指定国 (表示のない限り、全ての種類の国内保護が可能): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI,

(続葉有)

(54) Title: METHOD OF DETECTING OR QUANTITATIVELY DETERMINING MITOCHONDRIAL DNA 3243 VARIATION, AND KIT THEREFOR

(54) 発明の名称: ミトコンドリア DNA 3 2 4 3 変異の検出法および定量法ならびにそのためのキット



A...RELATIVE FLUORESCENCE INTENSITY (%)
B...CYCLE

(57) Abstract: A method of detecting a DNA having mitochondrial DNA 3243 variation, in which use is made of quantitative determination PCR using a primer having a base sequence complementary for a base sequence of 12 to 30 base length starting from base No. 243 in the base sequence of SEQ ID No. 1. Further, there is provided a method of detecting a DNA having mitochondrial DNA 3243 variation, in which use is made of a nucleic acid probe having its end labeled with a fluorochrome which upon hybridization exhibits a drop of fluorescence of the fluorochrome, the nucleic acid probe having a base sequence complementary for a base sequence of 14 to 40 base length starting from base No. 230 in the base sequence of SEQ IN No. 2, the nucleic acid probe having its 3'-end labeled with a fluorochrome.

(57) 要約: 配列番号 1 に示す塩基配列の塩基番号 243 から始まる 12~30 塩基長の塩基配列に相補的な塩基配列を有するプライマーを用いる定量的 PCR を用いる、ミトコンドリア DNA 3243 変異を有する DNA の検出方法、ならびに、末端が蛍光色素で標識され、ハイブリダイゼーション

(続葉有)

WO 2004/092415 A1

BEST AVAILABLE COPY



NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG,
SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ,
VC, VN, YU, ZA, ZM, ZW.

NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG,
CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

添付公開書類:

— 国際調査報告書

- (84) 指定国 (表示のない限り、全ての種類の広域保護が可能): ARIPO (BW, GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), ユーラシア (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), ヨーロッパ (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC,

2文字コード及び他の略語については、定期発行される各PCTガゼットの巻頭に掲載されている「コードと略語のガイダンスノート」を参照。

したときに蛍光色素の蛍光が減少する核酸プローブであって、配列番号2に示す塩基配列において塩基番号230から始まる14~40塩基長の塩基配列に相補的な塩基配列を有し、3'末端が蛍光色素で標識されている前記核酸プローブを用いる、ミトコンドリアDNA3243変異を有するDNAの検出方法。

INTERNATIONAL SEARCH REPORT

International application No.

PCT/JP2004/005496

A. CLASSIFICATION OF SUBJECT MATTER
Int.Cl⁷ C12Q1/68, C12N15/09

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
Int.Cl⁷ C12Q1/00-70, C12N15/00-90

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
JICST FILE (JOIS), EUROPAT (QUESTEL), MEDLINE/BIOSIS/WPIDS (STN)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X Y	P. SEIBEL et al., A Rapid and Sensitive PCR Screening Method for Point Mutations Associated with Mitochondrial Encephalomyopathies, Biochemical and Biophysical Research Communications, 1994, 200(2), p.938-42	1, 2, 8, 9 3-7, 10-24
X Y	M. ODAWARA et al., Selection of primers for detection of A to G mutation at nucleotide 3243 of the mitochondrial gene, Diabetologia, 1995, 38(3), p.377-8	1, 2, 8, 9 3-7, 10-24
X Y	C. ZHANG et al., Occurrence of a Particular Base Substitution (3243 A to G) in Mitochondrial DNA of Tissues of Ageing Humans, Biochemical and Biophysical Research Communications, 1993, 195(2), p1104-10	2, 9 3-7, 10-24

☒ Further documents are listed in the continuation of Box C.

☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search
02 July, 2004 (02.07.04)

Date of mailing of the international search report
20 July, 2004 (20.07.04)

Name and mailing address of the ISA/
Japanese Patent Office

Authorized officer

Facsimile No.

Telephone No.

BEST AVAILABLE COPY

INTERNATIONAL SEARCH REPORT

International application No.

PCT/JP2004/005496

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	JP 2002-119291 A (Japan Bioindustry Association), 23 April, 2002 (23.04.02), & WO 2002/008414 A1 & EP 1295941 A1 & US 2002/0106653 A1	3-7, 10-24
Y	K. TSUKUDA et al., Screening of Patients with Maternally Transmitted Diabetes for Mitochondrial Gene Mutations in the tRNA ^{Leu(UUR)} Region, Diabetic Medicine, 1997, 14, p.1032-7	16-24
A	J. LOEFFLER et al., Rapid Detection of Point Mutations by Fluorescence Resonance Energy Transfer and Probe Melting Curves in Candida Species, Clinical Chemistry, 2000, 46 (5), p. 631-5	1-24 08.4053.8 4054.8 3
A	Michizo NAKAMURA et al., "Hen'i Mitochondria DNA no Kenshutsuho no Shinpo mutation-specific PCR ni yoru mit DNA Ten Hen'i Kenshutsuho", 1997, 55 (12), p.3277-81	1-24
A	JP 11-221077 A (Otsuka Pharmaceutical Co., Ltd.), 17 August, 1999 (17.08.99), (Family: none)	1-24

BEST AVAILABLE COPY

INTERNATIONAL SEARCH REPORT

International application No.

PCT/JP2004/005496

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☒ Claims Nos.: (see below)
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
Some of claims 1-4 and 6-24 either fail to clearly define the scope of claims or are not fully supported by the description and are not clearly and fully disclosed in the description, so that any international search has been carried out. For the reason, see extra sheet.

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

Claims 1-15 are common to claims 16-24 in the technical matter of a method of detecting mitochondrial DNA 3243 variation. However, this common matter is publicly known as described in, for example, the following reference. Consequently, it cannot be stated that the claims 1-15 and the claims 16-24 share special technical features. Therefore, these claimed inventions do not constitute a group of inventions linked so as to form a single general inventive concept.

(Continued to extra sheet.)

1. ☒ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☒ No protest accompanied the payment of additional search fees.

BEST AVAILABLE COPY

INTERNATIONAL SEARCH REPORT

International application No.

PCT/JP2004/005496

Continuation of Box No.II-2 of continuation of first sheet(2)

G.S.P. YU et al., "Keiko Bio Image Analyzer (FM-BIO) ni yoru Mitochondria Idenshi 3243 Hen'I no Kenshutsu", The Japanese Journal of Clinical Pathology, 1996, 44(8), p.778-82

Claims 1, 3, 8, 10 and 12

With respect to the invention of these claims, taking into account that the base sequence of SEQ ID No. 1 is derived from wild type, a DNA having mitochondrial DNA 3243 variation cannot be detected by the use of a primer having a base sequence complementary for the above base sequence. Therefore, the invention of these claims cannot be stated as being fully supported by the description and are not clearly and fully disclosed to such an extent that an expert in the art to which the invention pertains can carry out the invention.

Incidentally, search has been conducted interpreting the primer according to the invention of these claims as a primer composed of a base sequence complementary for the base sequence of SEQ ID No. 2 derived from variant type, for example, a primer composed of the base sequence of SEQ ID No. 5.

Claims 1-4, 6-17, 22 and 23

With respect to the description "having a base sequence" used in these claims, whether or not "composed of a base sequence" is meant, "including a base sequence" is meant or anything else is meant thereby is ambiguous. Thus, these claims cannot be stated as being clearly drafted.

Likewise, in these claims, the wordings of "having a base sequence and" and "exhibiting a base sequence" are not clear.

With respect to the above ambiguous wordings, search has been conducted by interpreting them as meaning "composed of a base sequence" or "composed of a base sequence and".

Claims 16-24

Taking Examples, etc. into account, a nucleic acid probe capable of detecting a DNA having mitochondrial DNA 3243 variation is only a nucleic acid probe composed of a base sequence of SEQ ID No. 21 or 22. Except for this nucleic acid probe, what structure is had by the nucleic acid probe composed of "a base sequence complementary for a base sequence of 14 to 40 base length starting from base No. 230 in the base sequence of SEQ ID No. 2" usable in the detection is ambiguous. Therefore, the invention of these claims cannot be stated as being fully supported by the description and are not clearly and fully disclosed to such an extent that an expert in the art to which the invention pertains can carry out the invention.

No search has been conducted on the inventions other than the invention relating to the nucleic acid probe composed of a base sequence of SEQ ID No. 21 or 22, which are not fully supported by the description and are not clearly and fully disclosed in the description.

BEST AVAILABLE COPY